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THE INFLUENCE OF DIAPAUSE ON THE RESISTANCE TO DESICCATION OF EGGS OF *TELEOGRYLLUS COMMODUS* (WALK.) (ORTHOPTERA: GRYLLIDAE)

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Abstract

The eggs of two races of *Teleogryllus commodus*, one a non-diapausing race from a tropical latitude and the other from a temperate zone of Australia, were measured at each of two stages of development for resistance to desiccation. A comparison was also made between eggs in diapause and those at a similar stage of development from the same cricket cultures, from which diapause had been eliminated by prior treatment with ammonia.

It was found that eggs of both races were much more sensitive to desiccation prior to water uptake. The most resistant eggs were from the non-diapausing race after water uptake had been completed.

Diapausing eggs were more resistant than eggs of the same source from which diapause had been eliminated; but since eggs at a similar stage of development from a non-dispausing race were even more resistant, it would appear that the enhanced resistance may be incidental to, rather than a function of, diapause.

Introduction

One of the functions attributed to diapause is that in addition to being a means by which the life cycle is synchronized with the seasonal cycle, it confers on the diapausing organism 'an enhanced resistance to adverse climatic conditions such as cold, heat and drought' (Lees 1955). This implies that there are physiological changes accompanying the onset of diapause having the specific effect of rendering the organism more resistant to unfavourable conditions.

Despite the general acceptance of this viewpoint, there seems to be an absence of any reported experimental work demonstrating that organisms in diapause are more resistant than similar organisms in the same stage of development, but diapause-free. One reason for this is that direct evidence as to whether increased resistance due to physiological change is a concomitant of diapause is difficult to obtain in an organism with obligate diapause, since the most acceptable comparison is between diapause and non-diapause material tested contemporaneously. Thus the fact that in *Petrobia lutens* the winter diapausing egg has an additional layer of wax in the cuticle compared with the summer egg and is more resistant to desiccation (Lees 1954) does not constitute critical evidence, since presumably this could be a characteristic of the winter egg independently of diapause.

However, in *Teleogryllus commodus* it has been found that the application of ammonia under specified conditions eliminates diapause (Hogan 1964). It is possible, therefore, to compare eggs from the same cricket culture, and from the same egg batches, some of which have had diapause induced in them, and others from which diapause has been averted.

In this paper an experiment is described in which the relative capacity to resist desiccation of diapausing and non-diapausing eggs from the same source and at a similar stage of development is measured. As a further measure of the degree of resistance to desiccation in non-diapausing eggs, observations were made on the eggs of a non-diapausing race of *T. commodus* from northern Queensland. Finally, eggs of both races were tested at an earlier stage of development, prior to the stage at which water uptake occurs. This commences on the third day of development at 27°C, and is described by Browning (1953) who also studied the effect of desiccation.

Materials and Methods

Eggs from laboratory-reared progeny of crickets collected from the field were obtained by placing trays of moist sand into the cultures overnight and sieving out the eggs the next morning (Hogan 1965). Desiccation experiments were performed on batches of approximately 1000 eggs in each of the following categories.

(1) Eggs of the diapause race at the pre-diapause, pre-water uptake stage. These eggs had been incubated for three days at 23°C, commencing within eight hours of oviposition.

(2) Diapause eggs. Diapause had been induced by incubating the eggs at 23°C for a period of 12 days.

(3) Diapause-free eggs. Diapause had been averted by treating 3-day old eggs (at 23°C) with gaseous ammonia evolved from 100 mls. of 0·032M NH_4OH in a desiccator for a period of four days. The eggs were then washed and transferred on to water-soaked filter paper discs in plastic tubes and incubated at 18°C for a further four days and one day at 23°C, by which time water uptake had been completed in all the eggs. (Uptake of water appeared to be delayed to some extent by the ammonia treatment).

(4) Eggs of the non-diapausing northern Queensland race at the pre-water uptake stage similar to (1).

(5) Eggs of the northern Queensland race which had been incubated for eight days* at 23°C, by which time water uptake was complete. (The embryos would be more advanced than those of diapause eggs of the same age, which stop development just prior to water uptake).

* Water uptake takes place at this stage both in non-diapausing and diapausing eggs; but the latter require a further period to be in full diapause (Hogan 1960).

Desiccation

The desiccation of the eggs was carried out in a 10″ desiccator modified for the purpose by replacing the glass-domed top with a flat piece of perspex from which was suspended a 6″ × 4″ bronze gauze tray. This held small gauze wire trays in which the eggs for testing were placed. To enable these trays of eggs to be placed in and removed from the desiccator at the appropriate times, the perspex cover was fitted with a 2″ × 4″ opening covered by another piece of perspex sealed with petroleum jelly. This cover would slide aside when necessary with a minimum of disturbance to the air in the desiccator. Controls for each type of egg were held under the same conditions during the desiccation treatment but with water in the desiccators, and were then incubated on moist filter paper in tubes at 23°C to determine natural mortality.

The relative humidity was controlled by a sulphuric acid of SG 1·830 in the well of the desiccator. Variations in the R.H. were measured by means of a small, previously calibrated hair hygrometer suspended from the perspex cover. The desiccator was on a turntable rotating at 5 r.p.m. in a cabinet held at 18° (± 0·2°)C.

The trials were carried out at 18°C because at this temperature the rate of development of the eggs is very low and there would be little change in stage of development of eggs not in diapause during the period of treatment.

The more critical comparisons of the diapause, diapause averted, and non-diapausing eggs, were carried out synchronously on eggs of the same batch; eggs at the pre-water uptake stage were tested separately but under identical conditions to the other stages.

Three replicates of 50 eggs for each treatment were counted into the gauze wire trays and placed in the desiccator. One set of replicates for each type of egg was removed after 6, 16 and 32 hours and for types 2 and 5, also after 42 hours (see Section II). These periods of exposure to desiccation were based on preliminary experiments.

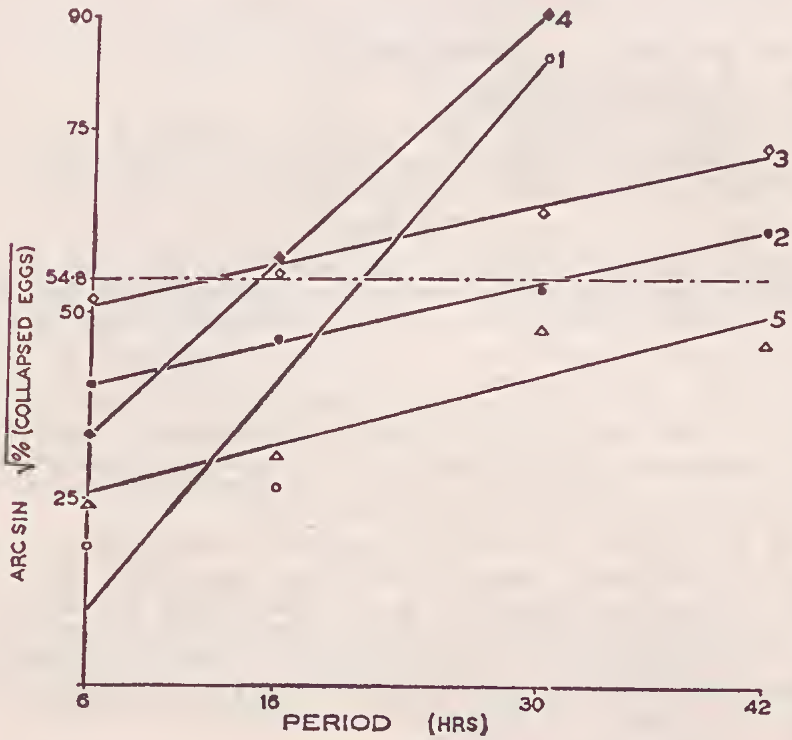

FIG. 1—The resistance to desiccation of five types of eggs of *Teleogryllus commodus* expressed as the arcsin percentage of eggs that collapsed in the period of exposure indicated on the abscissa. The types of eggs are as follows: (1) Pre-water uptake—diapausing race; (2) Diapausing (3) Diapause-free (4) Pre-water uptake, non-diapausing race (5) Water uptake, non-diapausing race.
For more precise details, see 'Materials and Methods', p. 38.

The effect of desiccation on the eggs at each interval of time was measured by counting in each group the number of eggs that completely collapsed as a result of the treatment. In previous tests all such eggs had been found to be dead when incubated at 23°C in the presence of moisture. Browning (1953) found that very few eggs were able to survive a 20 per cent loss by weight of moisture.

Results

During the course of the experiment, the R.H. varied from 1·5—5 per cent, the increases being associated with opening of the desiccator at the above intervals. Over the initial six hours, the mean R.H. was slightly lower than over the other periods but this applied to each type of egg and, if it has any significance at all, affects only the comparison between the effect of periods of treatment.

Fig. 1 shows that the stage of development was important in relation to the ability of the eggs to resist desiccation; the mean period for the collapse of the pre-water uptake eggs was significantly less than for the post-water uptake eggs of both the diapause and non-diapause races.

The diapausing eggs were significantly more resistant than diapause-free eggs of the same race, but these same diapausing eggs were significantly less resistant than the non-diapausing eggs, at a corresponding stage of development, from the Queensland race. It is clear, therefore, that the attribute of resistance to desiccation can exist quite independently of diapause.

In the control treatments the ammonia-treated Victorian eggs proved to have 20 per cent of the eggs in diapause. Presumably this would have the effect of giving this group a resistance to desiccation higher than if all of them were diapause-free.

Discussion

The greater susceptibility of the pre-water uptake stage of the eggs of both races to desiccation is not surprising if one considers the conditions under which this stage normally occurs. The mature females have the characteristic that they avoid ovipositing in dry soil and oviposition is usually delayed until adequate rain has fallen. Freshly oviposited eggs, therefore, are assured of an adequate moisture supply during the early stages of development. Adaptive selection for resistance to desiccation on pre-water uptake eggs would operate to a very limited extent, if at all.

Mature females of the Queensland race show the same aversion to ovipositing in dry sand when reared in laboratory cultures. Presumably, water uptake, in the field, is assured in the same way as in the Victorian race.

At the other extreme, eggs from the Queensland race, when water uptake is complete, are much more resistant to desiccation. Again, this appears to be in accordance with the conditions they may experience. Should drought occur in the tropics, then it would be expected that the prevailing high temperatures would lead to severe desiccating conditions. Victorian eggs at this stage of development may experience drought conditions too, but the temperatures would be lower and in the late autumn, cool nights would lead to condensation of moisture from the air so that conditions would be relatively moderate.

When comparing the resistance to desiccation of the pre-water uptake eggs and the post-water uptake eggs, it must be taken into account that the former have a smaller volume, less water and a relatively larger surface area. Hence, if the permeability of the cuticle were the same in both types of eggs, those at the pre-water uptake stage would be expected to collapse more rapidly.

The results obtained with the non-diapausing race from northern Queensland demonstrate that adaptation for resistance to desiccation in eggs of *T. commodus* is possible without involving diapause, and this provides some evidence to support the idea that the function of diapause in the Victorian eggs is primarily to synchronise the life cycle of the insect with the climatic cycle of the seasons.

Nevertheless, the foregoing results show that when diapause is eliminated, eggs of the Victorian race are more susceptible to desiccation. It is apparent, therefore, that there must be physiological changes in the eggs accompanying the entry into diapause that confer increased resistance to desiccation. Since, however, resistance to desiccation is high in eggs of the non-diapausing race, it is concluded that the extra resistance associated with diapause is incidental to, rather than a function of, diapause.

Salt (1961) has reached a similar conclusion in relation to cold hardiness and diapause. In his opinion, cold hardiness due to the accumulation of glycerol in the blood, though acquired concurrently with the entry into diapause, is not causally related to it.

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